

IN THE SPECIFICATION:

Please replace the paragraph beginning on page 2, line 6 and ending on page 2, line 12 with the following:

MAL was obtained by passing a casein fraction of milk, particularly human milk, down an ion exchange column, specifically a ~~DEAE-Tris-acyl~~ Diethylaminoethyl ("DEAE")-[tri-hydroxymethyl-aminomethane] ("TRIS")-Acryl M column using an NaCl gradient. A pool containing MAL which was active therapeutically was obtained. In addition, the conversion of commercially available monomeric α -lactalbumin to the active form was reported.

Please replace the paragraph beginning on page 4, line 32 and ending on page 5, line 6 with the following:

The A-state or molten globule state of α -lactalbumin has native-like secondary structure but less well-defined tertiary structure (Kronman et al. 1965 Biochem, 4, 518-525; Dolgikh et al. Febs Lett, (1981) 136, 311-315 and FEBS Lett, (1984) 165:88-92, Ohgushi & Wada, 1983, A Febs Lett, 164:21-25). Molten globules are formed under acidic conditions and similar states are formed at neutral pH upon removal of the tightly bound Ca^{2+} -ion by ethylene diamine tetraacetic acid ("EDTA") ~~EDTA~~ by reduction of the disulfide bonds, or at elevated temperatures (Pfeil et al., 1987 Biochim Biophys Acta, 911:114-116; Kuwajima 1996 Faseb J. 1:102-109; Shulman et al., 1995 J. Mol. Biol. 253, 651-657).

Please replace the paragraph beginning on page 5, line 32 and ending on page 6, line 5 with the following:

Depending upon the purification process or the source of the α -lactalbumin used in its production, it may be preferable to subject the α -lactalbumin to a pretreatment step which maximise amount of molten globule-like material. This may be effected by contacting the α -lactalbumin with a calcium chelating agent such as EDTA (~~ethylene diamine tetraacetic acid~~) in order to remove excess calcium. This may be applied as a pre-treatment in which the α -lactalbumin is contacted with the chelating agent prior to elution down the ion exchange column, or alternatively, the EDTA may be added to the elution buffer.

Please replace the paragraph beginning on page 8, line 31 and ending on page 8, line 34 with the following:

Suitably the oleic acid or casein or the active components thereof are eluted through a column containing new unused ion exchange material such as DEAE-TRIS-Acryl ~~DEAE-Trisacryl~~. Suitable elution buffers include tri-hydroxymethyl-aminomethane hydrochloride ("TRIS-HCl") ~~Tris-HCl~~ with a pH of 8.5.

Please replace the paragraph beginning on page 9, line 9 and ending on page 9, line 15 with the following:

In a preferred embodiment, the column is then washed with ion exchange buffer, such as the ~~Tris-HCl~~ TRIS-HCl buffer mentioned above, without casein, and preferably also other buffers which are to be used in the process, such as a NaCl containing buffer to ensure that nothing unspecific will elute from the column when used in the process. Washing may be done several times.

Please replace the paragraph beginning on page 12, line 11 and ending on page 12, line 15 with the following:

Methodology

The ion exchange matrix used in the following examples was ~~DEAE-Trisacryl~~ DEAE TRIS-Acryl M from BioSeptra, France. The buffers comprised: Buffer A, 10mM ~~Tris-HCl~~ TRIS-HCl pH 8.5 and Buffer B, 10 mM TRIS-HCl ~~Tris-HCl~~ with 1M NaCl pH 8.5.

Please replace the paragraph beginning on page 13, line 1 and ending on page 13, line 4 with the following:

Product was analysed using gel electrophoresis ~~Tris-Glycine~~ TRIS-Glycine PAGE gels 4-20%) as described previously in WO 96/04929, the content of which is hereby incorporated by reference.

Please replace the paragraph beginning on page 16, line 36 and ending on page 17, line 23 with the following:

Frozen human milk was thawed and centrifuged (Sorvall RC-5B refrigerated superspeed centrifuge, Du Pont Instruments, Wilmington DE, USA) at 2500 x g for 15 minutes; the upper fat layer was removed. Casein was isolated by an overnight incubation at +4°C with 10% potassium oxalate followed by a second overnight incubation at +4°C after lowering the Ph to 4.3 using 1 M hydrochloric acid and heating the solution to 32°C for 2 hours. The casein precipitate was harvested by centrifugation at 5000 x g for 15 minutes, washed by 3-5 cycles of certrifugation and resuspension in distilled water and lyophilized. Casein was further fractionated on an ion-exchange column (14 cm x 1.5 cm) packed with ~~DEAE-Trisacryl~~ DEAE-TRIS-

Acryl M (BioSeptra, France) attached to a Biologic chromatography system (Biorad laboratories, Alfred Nobel Drive, Hercules, CA, USA) using an increasing gradient of NaCl. The run was under the following conditions: buffer A: 10 mM TRIS-HCl ~~Tris/HCl~~ pH 8.5; buffer B: buffer A containing 1 M NaCl/L. Gradient program: From start to 15 ml, 0% B; from 15 to 55 ml, 0-15% B; from 55 to 75 ml, 15% B; at 75ml, 100% B for 10 min; from 85 to 115 ml, 0% B; at 115 ml, 100% B for 20 min; from 135 to end 0% B. The flow rate was 1 ml/min and the fraction size was 0.5 ml. The peaks was monitored by absorbance at 280 nm. The elute was desalted by dialysis (Spectra/Por, Spectrum Medical Industries, Laguna Hill CA, USA, membrane cut off 3.5 kD) against distilled water for at least 48 h and lyophilized.

Please replace the paragraph beginning on page 17, line 36 and ending on page 18, line 4 with the following:

Size exclusion chromatography was effected by gel filtration, performed on a Pharmacia Sepharose 12 (S-12) gel filtration column in 10mM TRIS-HCl ~~Tris/HCl~~ pH 7.5 with 0.15M NaCl, and monitored by UV absortion at 280nm. The flow rate was 0.3 ml/min and the fraction size was 0.5ml. Observed peaks were collected and desalted by dialysis against distilled water.

Please replace the paragraphs beginning on page 20, line 21 and ending on page 21, line 4 with the following:

Example 6

Conversion of Monomeric α -lactalbumin to MAL

Monomeric α -lactalbumin was purified from human milk by ammonium sulphate precipitation. The ammonium sulphate was added as a salt, 264g/1 milk, and the

mixture was incubated overnight at +4°C. The mixture was then centrifuged (Sorvall RC-5B refrigerated superspeed centrifuged, Du Pont Instruments, Wilmington DE, USA) at 5000 x g for 15 minutes. The whey fraction was collected, lyophilized and dissolved in 50 mM ~~Tris/HCl~~ TRIS-HCl with 35 mM EDTA, pH 7.5. A 400 ml phenyl-sepharose column (Pharmacia Biotech, Björkgatan, Uppsala, Sweden) was packed in 50 mM ~~Tris/HCl~~ TRIS-HCl with 1 mM EDTA, pH 7.5 and 500 ml sample was loaded onto the column. The column was first eluted with 50 mM ~~Tris/HCl~~ TRIS-HCl with 1 mM EDTA, pH 7.5 and α -lactalbumin was then eluted from the column with 50 mM ~~Tris/HCl~~ TRIS/HCl with 1 mM CaCl_2 , pH 7.5.

For conversion of monomeric α -lactalbumin, samples were dissolved in 10 mM ~~Tris/HCl~~ TRIS-HCl pH 8.5 (A buffer) prior to loading onto the column. Material that eluted at 1 M NaCl was collected, desalted by dialysis and lyophilized.